IN THE UNITED STATES DISTRICT COURT

FOR THE NORTHERN DISTRICT OF OKLAHOMA

STATE OF OKLAHOMA, ex rel, W.A. DREW EDMONDSON, in his capacity as ATTORNEY GENERAL OF THE STATE OF OKLAHOMA, et al.	)	
Plaintiffs,	)	
V.	)	No. 05-CV-329-GKF-SAJ
TYSON FOODS, INC., et al.,	)	
Defendants.	)	

REPORTER'S TRANSCRIPT OF PROCEEDINGS

FEBRUARY 20, 2008

PRELIMINARY INJUNCTION HEARING

VOLUME II

BEFORE THE HONORABLE GREGORY K. FRIZZELL, Judge

APPEARANCES:

For the Plaintiffs: Mr. Drew Edmondson
Attorney General
Mr. Robert Nance
Mr. Daniel Lennington
Ms. Kelly Hunter Burch
Mr. Trevor Hammons
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313 N.E. 21st Street
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work that we do.

Q. Well, let's back up because maybe I misunderstood.

MR. BULLOCK: Judge, we're well past the half hour, I just wonder when counsel is going to wrap up. I'm not trying to hold people to specific --

MR. GEORGE: Two minutes, Your Honor.

THE COURT: Very good.

Q. (By Mr. George) I want to make sure I understand, Dr. Teaf. You're not offering an opinion in this case regarding the likelihood of transport of poultry litter to a water body compared to other sources; is that correct?

A. No, I'm not. No, I'm not. I'm identifying sources, and I'm identifying receptors.

Q. In fact, yesterday when you talked about -- I think you threw out some percentages in terms of cattle manure versus poultry litter. You were talking just about your analysis of how much hits the ground, not how much gets to the water; correct?

A. And subsequent to that I discussed the importance of knowing how it may make its way to the water body, yes, sir.

Q. But you're not offering an opinion as to whether it got there or not because you're not offering a fate and transport opinion; correct?

A. Well, I am offering an opinion about that it got there and I'm offering it for two reasons. One, the bacteria levels are

very high and second of all, the signature that was identified is of cattle -- is of poultry.

Q. You're relying upon the work of Dr. Roger Olsen for your belief that the water shows the evidence of poultry contamination; correct?

A. In part I am and I'm also relying upon that of Dr. Harwood and the other lines of evidence that I described yesterday.

Q. But you yourself, sir, have conducted no fate and transport analysis; correct?

A. No, I did not, not a formal one, no.

Q. Sir, based upon the work that you've done in this case, not the work of others, can you state to a reasonable degree of scientific certainty that if Judge Frizzell grants the injunction that is requested by your client, the water quality standards for bacteria in the Illinois River will be met in 2008 and 2009?

A. My opinion is that they will be.

Q. Can you state that opinion to a reasonable degree of scientific certainty?

A. I can based on the information that I have reviewed.

Q. You're willing to stake your professional reputation on the proposition that if this Court enters the injunction sought by your client, the water quality standards for bacteria in the Illinois River will be met next year?

A. Based on all the information that I have and my knowledge

A. Yes, there is. And the reason that I just didn't recall at the time -- the Wise County cases involved bacterial growth producing hydrogen sulfide in residential wells as a consequence of the introduction of natural gas and condensate. So I didn't think about them as coming from the surface, but the contaminant of concern was hydrogen sulfide is microbially produced.

Q. Sir, you were not asked to evaluate in that case the fate and transport of bacteria found in groundwater, were you?

A. No.

Q. You were simply evaluating the effects of groundwater -- I'm sorry, of bacteria found in certain wells?

A. That's correct.

Q. So as it stands today, sir, you have never before worked on a litigated matter in which you were asked to offer an opinion as to the fate and transport of bacteria to groundwater?

A. That's correct.

Q. Sir, prior to being retained by the Plaintiffs' lawyers representing the attorney general's office in this case, had you ever worked on a research project or published a paper related to the movement of bacteria in either surface water or groundwater?

A. No.

Q. Sir, have you ever had your opinions in an environmental

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FEBRUARY 21, 2008

PRELIMINARY INJUNCTION HEARING

VOLUME III

BEFORE THE HONORABLE GREGORY K. FRIZZELL, Judge

APPEARANCES:

For the Plaintiffs: Mr. Drew Edmondson
Attorney General
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Mr. Daniel Lennington
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as a reliable method of tracking fecal bacteria in the environment?

A. Yes, as I said, they have several experts working on this area themselves.

Q. Dr. Harwood, I'd like to call your attention to State's Exhibit 59-1. It should be in front of you there on the lectern in front of you.

A. Yes.

Q. Would you please identify that for the record?

A. Yes, that's my CV.

Q. Is it a current copy of your curriculum vitae?

A. Yes, it looks like it.

Q. Have you recently updated that curriculum?

A. Yes, just recently we had a paper that's been published in Applied Environmental Microbiology on quantitative PCR, so that was an updated edition.

Q. You said quantitative PCR?

A. Quantitative polymerase chain reaction.

Q. So PCR stands for?

A. Polymerase chain reaction.

Q. I'm going to let you say that all day, I'm going to say PCR.

A. Okay. Me, too.

Q. When did you first become involved in the cases before the Court here today?

A. I was first contacted in August 2004 and then did not start working on the case until April 2005.

Q. Now, what is your understanding, Doctor, about the subject matter of the case that's before the Court today?

A. The Oklahoma Attorney General has filed suit against some poultry integrators in order to stop or place a moratorium upon land application of poultry litter due to environmental, ecological and human health hazards associated with that practice.

Q. Were you given any assignments in this case?

A. I was asked to help plan sampling procedures, review analytical results for microbiology analyses and render opinions on the -- on aspects of microbiological water contamination from land applied poultry litter and human health risks that could result from that practice. And also worked in conjunction with North Wind Laboratory to develop what we term a poultry litter biomarker, a specific PCR assay for bacteria that are associated with poultry litter, to use as a tracer for land applied poultry litter.

Q. Okay, Doctor. Doctor, what materials have you reviewed in order to accomplish those assignments?

A. Well, I've reviewed a lot of documents, but they include results of microbial testing that were sent to me by CDM. And the analyses were done by laboratories, three laboratories, FoodProtech, A&L Laboratory and EML Laboratory. I reviewed

represented by them.
Q. Thank you, Doctor. I want to switch gears on you a little bit again. Do you have an opinion with respect to the source of bacteria that has impaired the IRW?
A. Yes, I believe that a significant portion of that is contributed by land applied poultry litter.
Q. And do you have an opinion as to what would happen if poultry waste land application was stopped in the IRW?
A. Yes, I believe that over time the levels of bacteria would decline and that the human health risk would be decreased.
Q. Okay. Do you have any specific evidence, Doctor, that contribution of poultry litter to lands in the IRW has contaminated the waters of the IRW?
A. Yes, we used a reliable method called polymerase chain reaction or PCR to develop a poultry litter specific biomarker which we use as a tracer to follow the pathway of poultry -- of microbial contamination from poultry litter throughout the Illinois River Watershed.
Q. Would you just define briefly what a biomarker is?
A. A biomarker would be a biological component of some organism. In this case it's a bacterium and in this case the biological component is a gene fragment that we were able to detect by PCR and this bacterium is highly associated with chicken -- with contaminated chicken litter.
Q. Doctor, are there differences between the PCR method of

identification and then the standard methods of identifying bacteria such as the indicator bacteria?

A. Yes, as I mentioned before, standard methods for the last century have relied upon culturing bacteria. By the last century, I mean 1900 to 2000. Culturing bacteria is time consuming. Again, as I mentioned, it depends on the physiology of the bacteria, whether they were in a state to be able to grow or not and requires that one use the correct medium, that one has the correct incubation temperature. So culture-based methodologies are fraught with difficulties of interpretation.

PCR-based methods are basically being able to detect a specific genetic component of the bacterium. We use DNA -- we use the PCR as sort of a DNA Xeroxing machine. It's highly specific. It can amplify or produce large amounts of DNA from small amounts. It's rapid and it doesn't depend on the physiological state of the organism for detection. And again, it's actually much more highly specific than culture-based methods for bacterial identification are.

Q. Now, is PCR considered by the scientific community to be a reliable method to detect specific bacteria?

A. Yes, and in other scenarios other than bacterial use or identification of bacteria as well. So it's used, for example, in the legal field to determine the guilt of criminals or to free innocent people. It's also used in the medical setting to, again, to -- this goes back to the bacterial component --

to identify bacteria, viruses and other infectious microorganisms that cause disease. It's very widely used in the forensic and the clinical communities and it's making major inroads into environmental microbiology as well.

Q. So is your testimony, Doctor, that the PCR method that you employed in this case is the same methodology that's used to look at DNA in the criminal context to determine whether someone's DNA is in a crime scene or something like that?

A. It is essentially the same type of methodology.

Q. Is it the same methodology they use in hospitals to identify the source of a disease?

A. Yes, essentially the same.

Q. Okay. Now, Doctor, are you aware of a standard conventional method of detecting poultry bacteria in environmental media?

A. There is no standard conventional method for specifically detecting poultry contamination in environmental waters.

Q. So when you are faced with a hypothesis as an environmental question like this, how do you go about answering the question of such hypothesis?

A. Well, that's one of the things that my laboratory specializes in is developing methodology that can be validated in controlled settings and then used in the field to answer questions about where microorganisms come from in waters.

Q. Is that what you did when you developed the PCR

methodology in this case?
A. Yes, it is.
Q. Doctor, I want to call your attention to State's Exhibit 435. And again, there's a copy in the packet in front of you but there's also a blow-up of the exhibit on the tripod. Would you identify this document for the record, please?
A. Yes, this is a chart that shows -- that outlines the development and validation of the poultry litter biomarker for this study.
Q. Who prepared this exhibit?
A. This exhibit was -- well, the flowchart was prepared by myself.
Q. Okay. Would you take a couple of minutes and explain to the Court the methodology that you employed to develop the PCR biomarker in this case using this exhibit?
A. Yes, so to start off --
Q. You can stand up if you like or you can sit there with a pointer, either way.
A. I think I'm good here, that way everybody can hear me.
Q. Okay, thank you.
A. Keep in mind that what this -- the end goal of this process is have some sort of a genetic tracer that we can use to determine whether poultry litter was present in environmental samples, whether it be soil samples or water samples, groundwater, surface water. And so in order to do

that, we needed to find a genetic -- a piece of genetic material that came from microorganisms from the chickens. And it needed to be both specific to the poultry, broadly distributed in the waste, the poultry waste, and in field samples to which these -- this litter had been land applied. So it needed to be broadly distributed and also needed to be specific to the poultry contamination source. So that's the end gain.

The starting material we used to find this fragment, because keep in mind, none existed -- not none was existed, but none was identified before this process, was we used litter samples from poultry houses that contained chickens and those that contained turkeys and we used samples from fields to which poultry litter had been land applied. We --

Q. Is this all IRW based litter and fields?

A. This is all material from the IRW, yes.

Q. Thank you, Doctor.

A. It's all material from the IRW. We utilized polymerase chain reaction and we used three separate PCR, polymerase chain reaction, assays using what we call different primers. Primers are like little sticky bits of DNA that are very specific to the sequence that you're trying to amplify or make more of. And we used these -- and the PCR primers allow one to be very, very specific in terms of the genetic material that you are targeting. So we used separate PCR assays and separate primer

sets to develop a pool of E. coli DNA. So in one sample of poultry litter, for example, you might have ten or a hundred or even more different E. coli strains. So this DNA pool contained amplified or PCR amplified E. coli DNA. A second pool contained DNA from bacteria. And a third pool contained DNA from bacteroides. This is a fecal anaerobe that's been used in many other microbial source tracking studies.

We then used a method called terminal restriction fragment length polymorphism. This is basically going to cut the DNA depending on its precise sequence and give us fragments of variable lengths. And what we were looking for from these DNA pools were fragments that comprised at least 20 percent of the total DNA in the pool and that also were found across all of these samples because a biomarker that's infrequently found in the sample type is not going to be very useful once it gets out in the environment. It simply won't be present at high enough concentration and it won't be useful for a lot of different samples.

Q. Doctor, let me just ask you here. So on the right-hand side about a quarter of the way down you have criteria, unique poultry gene found in all samples. Is that what you just described in simple terms?

A. Right, that's what I just described. We're looking for a unique -- a gene that's unique. And it should actually say unique poultry bacteria gene because we're not really looking

for a gene from the chicken. We're looking for a gene from the bacteria associated with the chicken found in all of these samples because, again, we want it to be representative broadly of litter and land applied field samples.

Q. Thank you, Doctor. Please proceed.

A. All right. So we identified some candidate fragments from the TRFOP, the terminal restriction fragment length polymorphism, that were broadly present in these samples. And then we needed to further investigate these fragments because I said that the fragments needed to be broadly distributed that we're going to look at, but they also needed to be specific to poultry. And so we cloned these fragments. We did DNA sequences, so we determined their exact sequence. And then we matched the sequence of those fragments up to the GenBank database. This is a world-wide database containing literally millions of DNA sequences.

What we were looking for in the matching to the GenBank database was we were looking for fragments, for DNA fragments that have never been seen before in any other type of fecal material or in uncontaminated soil samples or in river water. We were basically looking for bacteria that are candidates for being poultry litter specific. And so what we found after this analysis, we submitted a lot of sequences to the --

MR. JORGENSEN: Your Honor, before we get to what we

found, I've been trying not to interrupt but I think it might be the right time. I know this is not a jury case and that there is no Daubert hearing. Just for the record, I want to say that we're going to make one. Dr. Harwood just testified that she -- no one has done this before, she invented this process. Obviously I suspect you would rather for me to wait and do it all on cross and then make it at the end. But I just, for the record, before the conclusion is stated, I want to say that we're going to say that this could never meet the standards of Kumho Tire.

THE COURT: Yes, sir, I understand that. And it appears that everyone is seeing it the same way procedurally as I am. Obviously Daubert is used to try to keep junk science away from juries. Obviously with a judge, I can make that determination. Your objection has been made for the record. Go ahead, Mr. Page.

MR. JORGENSEN: Thank you, Your Honor.

MR. PAGE: Thank you, Your Honor.

Q. (By Mr. Page) Dr. Harwood, I think you were talking about developing new PCR primers?

A. That's correct. So based on the --

Q. Just to ask a question, is that what you typically do with this type of work?

A. Yes, that is a strategy that has been employed in developing several of the most successful microbial source

tracking markers that are utilized.

Q. Would they develop these type of primers if they are doing work for a criminal case or a hospital analysis?

A. For hospital analysis, yes.

Q. Thank you, Doctor. Continue.

A. So we were -- after analyzing many different fragments and determining that some of these fragments were found in environments or fecal samples that were not from poultry litter, we ended up with three candidate primers for -- three candidates fragments that could possibly be a good biomarker. So we developed new PCR primers to make PCR assays for these candidate markers. With our new PCR assays in hand, we then went back to the litter samples and to the land applied field samples and made certain that we could amplify these targets out of these original samples. And we had subsamples of these samples. So again, we're making sure that this is broadly distributed in these so-called target samples.

Q. Would you explain what you mean by amplify?

A. Yeah.

Q. What does that look like?

A. So amplify, in PCR, when -- you start off with a very, very small amount of material and you use -- of genetic material, DNA. And you use the polymerase chain reaction as a -- again in colloquial terms you might say sort of a DNA Xeroxing machine to specifically increase the number of copies

of that particular piece of DNA that you're looking at.

Q. Thank you.

THE COURT: Let me go back just so I understand your flowchart. You say you developed three candidate primers here?

THE WITNESS: Right.

THE COURT: Where are you referring, the middle where it says three candidate genes? You've got another one over here, one candidate gene, which I think is four. What are you referring to here?

THE WITNESS: This here?

THE COURT: No, when you refer to you developed three candidate primers.

THE WITNESS: Oh, sorry, yes, there are four. This is three candidates from the DNA pool and one candidate from the E. coli pool. I just misspoke, sorry.

THE COURT: Okay.

THE WITNESS: So it would be four candidate biomarkers and four different PCR assays. So again, you needed to make sure that these PCR assays would work and amplify the fragment from our starting material. We also needed to go to the feces of animals that might -- whose fecal litter might impact the watershed or that might cross-react with this marker. So we collected numerous samples from cattle, from swine, from humans, both from wastewater treatment plants and from septic tanks, and from ducks and geese. And we utilized these PCR

primers and PCR assays against all of these so-called non-target samples.

And in fact, we had no cross-reactivity with the samples with the exception of one duck sample and one goose sample that came from outside of the Illinois River Watershed. We did have cross-reactivity of the marker with those samples. And then when we sequenced that PCR product, that bit of DNA that was Xeroxed or amplified during the process, we did find that it was the same DNA sequence as our biomarker. So we do have to accept some limited cross-reactivity in those reactions. However, that is the case with all microbial source tracking markers or the vast majority of them that have been found to date.

Q. (By Mr. Page) How does that cross-reactivity with that one duck and goose sample affect the reliability in your opinion of the PCR identification?

A. So it depends on the context that it's done in, but one of the aspects of this detection was for both the duck and the goose sample, we had duplicate samples. And only one of the duplicates -- in only one of the duplicates was amplification ever obtained which means -- and when we cloned those DNA sequences, we found a very low frequency within the DNA pool that we developed. So these sequences are present at very low concentration in the duck and goose feces and, again, infrequently. So as I said before, it will be a very poor

biomarker for duck and goose feces and, of course, a much, much better one for poultry feces for which the assay was derived.

THE COURT: But if I'm to understand correctly, it is a marker for duck and goose feces as well as general poultry feces?

THE WITNESS: It could pick up contamination from duck and goose feces. And so the -- what one has to do then in the weight of evidence approach that we use in these studies is to ascertain the extent to which duck and geese are present in the watershed to which they might be contributing contamination.

Q. (By Mr. Page) Would you compare that to the amount of poultry that are in the watershed?

A. In terms of numbers?

Q. And the weight of evidence, yes, in terms of numbers.

A. Oh, in terms of the weight of evidence, yes. So there's vastly, of course, vastly more poultry in the watershed than there are ducks and geese.

So after all of these -- after all of this development to date and validation, we were left with one primer set. We call the primer set LA-35 but I probably won't say that again. But LA-35 amplifies a DNA fragment from a bacteria that's most closely related to Brevibacteria avium. Brevibacteria avium is a bacterium that was first isolated from poultry. To my knowledge, it has not been isolated from organisms other than poultry. And so it appears to have a good basis for -- a good

biological basis even for being a poultry specific marker.

What we felt we needed to do at that point was --

Q. So at this time, at this point in the analysis, has the analysis been able to specifically identify poultry versus non-poultry feces bacteria?

A. Right. So at this point then we are able to go back to these field samples and utilize the PCR method on them. We've also been able to go to many other litter samples and edge of field samples and surface water samples and also validate that this PCR method works on -- broadly on poultry litter samples.

THE COURT: Excuse me for missing it, but how did we get from the four PCR primers to PCR LA-35? How did you make that --

THE WITNESS: So the other three, the other three primer sets either were deficient at amplifying from the original samples or, and this was even more the case, cross-reacted strongly with some of the components of our non-target samples. So we basically discarded them because they weren't useful as biomarkers.

THE COURT: All right. So LA-35 was one of the four?

THE WITNESS: Yes.

THE COURT: Okay.

THE WITNESS: LA-35 would have been up here in this box as a candidate gene from the DNA bacterial pool. And so it's made it all the way down here now.

THE COURT: All right.

THE WITNESS: The quantitative PCR was developed because at this stage, the LA-35 stage, this is a conventional PCR technique and it doesn't give us any numbers. It just gives us presence, absence. And we knew that we'd have additional information about the pathway of poultry contamination in the watershed if we were able to have a quantitative assessment of the amount of this biomarker that was finding its way into the water. And so the quantitative PCR assay was developed again based on these LA-35 primers using the same primer set. And we found it to be highly quantitative and sensitive with a detection limit of about six gene copies of the Brevibacteria avium-like bacteria. We don't know that this bacterium is Brevibacteria avium, but it is similar to that organism.

We then carried out the -- the qPCR means quantitative polymerase chain reaction. We carried out the qPCR analysis on various types of samples, including litter, soil, edge of field samples. Edge of field means the runoff that's coming directly off of the land applied fields. Also on surface water and groundwater samples.

THE COURT: What do you mean by detection limit of six gene copies?

THE WITNESS: That's what we call a method detection limit. So that means that the smallest amount of DNA that we

can detect from inside bacteria is six gene copies. That's called the sensitivity of the reaction.

THE COURT: Go ahead.

MR. PAGE: Thank you, Your Honor.

Q. (By Mr. Page) So at this point in time then you've been able to identify a method to specifically identify whether there's poultry bacteria genetic material in an environmental media?

A. Correct.

Q. And you are also able to quantify the relative amount?

A. Correct.

Q. Can you quantify in all samples?

A. In all sample types?

Q. No, on all samples where you find presence of the bacteria genetic material.

A. Well, it turns out that as you -- the way that we do this analysis, when we filter water and try to detect the bacteria, we have a detection limit of about six, of about -- sorry. We have a quantitative detection of about six -- 2,000 copies per liter of water, sorry. So for it to quantitate the biomarker, we need about 2,000 copies per liter of water. In order to simply detect the bacteria, we need more like 50 copies per liter of water. And the reason that that 50 number is different from the six is because we have to concentrate that sample and we have to extract the DNA and we're always losing a

little bit of sensitivity in that process.

Q. Thank you, Doctor. Who did you work with in development of this PCR process?

A. I worked with North Wind Laboratory and that was Tamzen Macbeth and Jennifer Weide were the scientists there that I worked with.

Q. Anyone else?

A. We worked with Roger Olsen in terms of we worked on the sampling strategy and collection.

Q. Do you intend to publish your findings of this study in a peer reviewed scientific journal?

A. Yes, definitely. The abstract is submitted to the American Society of Microbiology Conference which will take place in June. And the manuscript is in preparation to be submitted to Applied Environmental Microbiology.

Q. Doctor, now I want to turn your attention to Plaintiffs' Exhibit 436.

THE COURT: Doctor, I imagine this will be touched upon in cross-examination, but to the extent the manuscript is in preparation, it hasn't been subjected to peer review or scrutiny; correct?

THE WITNESS: Correct.

THE COURT: Go ahead.

MR. PAGE: Thank you, Your Honor.

Q. (By Mr. Page) Dr. Harwood, would you please identify for

MR. PAGE: Thank you, Your Honor.

Q. (By Mr. Page) Did you detect the biomarker in surface water samples?

A. Yes, we did. We detected the biomarker in 43 and a half percent or so of surface samples at levels up to 100,000 per liter.

Q. What about groundwater samples?

A. We did detect it in some groundwater samples, two groundwater samples to be exact, and at a level up to 20,000 per liter. And two out of 22 samples would be 9 percent.

Q. Now, a similar question to what the Judge just asked you. What does this information tell you, if anything, with regard to the distribution or pathway of poultry waste bacteria in the IRW?

A. Well, it demonstrates that the bacteria are following the pathway or that they have a transport pathway from the fields to the surface waters and also into the substratum into that karst, that fractured karst substratum which then allows them to appear in the groundwater and then be transported back upward into the spring systems.

Q. Let me draw your attention or if you would, to sample marked LAL5A on this exhibit. Can you identify that location for the Court, please?

A. Yeah, I think so. LAL5A is right about here. That's a soil sample and from a land applied field. That one had 4

break?

MR. PAGE: I would, Your Honor, thank you.

THE COURT: Let's take a recess until how's 1:30? Is that enough time? We'll be in recess until 1:30 p.m.

(Recess.)

MR. PAGE: Your Honor, thank you for calling that break. May I continue, Your Honor?

THE COURT: Yes, sir.

MR. PAGE: Thank you, sir.

Q. (By Mr. Page) Dr. Harwood, how many samples have been analyzed for PCR to date?

A. A little bit over -- a little bit over 200.

Q. And how many total samples are there?

A. About 550.

Q. And how come your analysis ends with 200 samples?

A. We had -- we received results of the sampling in October, November and January. And after that, we were instructed to stop submitting new results until after this hearing is my understanding.

Q. Thank you. I'd like to turn your attention to Exhibit 439. Dr. Harwood, can you identify State's Exhibit 439?

A. That is a graph that was prepared under my direction. And it shows on the vertical axis -- well, it's a comparison of the results for the poultry biomarker assay versus the concentration of Enterococci in various samples, including

litter, soil, edge of field, surface water and groundwater samples.

Q. What does this graph tell us with regard to a relationship between the bacteria that are shown on it?

A. Well, it tells us a couple of things. First of all, there is a significant relationship between Enterococcus concentrations and the concentration of the poultry litter biomarker in these samples. It also tells us something else. We talked about the sensitivity of the assay and how much needed to be present to be quantified and so you need about 2,000 copies of the gene to quantify. And when I prepared this graph, what I did was I used the quantitative results for this cluster. But if a sample had presence of the biomarker, but it was not enough to quantify, then I assigned it a value of one. So that's those values down here. And then if the biomarker was not present, I assigned a value of zero. So that's what these are right here.

But even though we do have this gap in the ability to quantify in this area, we still do have a strong correlation between Enterococci and the Brevibacterium poultry litter biomarker. And you see here the P value is .0001 which means that there is only one chance in a thousand that this relationship between the variables is occurring by chance.

Q. Does it tell us anything about the relationship between poultry waste and the Enterococci indicator bacteria we're

finding in our samples?
A. Well, it does say that they co-occur. So when you tend to have high levels of Enterococci, you also tend to have high levels of the biomarker.
Q. Thank you. Now, let me show you Exhibit 438.
A. That's a very similar graph except that shows the relationship of the biomarker, the poultry litter biomarker, with E. coli concentration. And it's another indicator bacteria that we're using for general fecal contamination.
Q. Again, does it indicate anything with regard to the relationship between the E. coli that's found in the environment and the PCR Brevibacterium?
A. Well, again, when we have high levels of E. coli, we also tend to have high levels of the Brevibacterium.
Q. Thank you. And then again, let me show you what's been marked as Exhibit 440.
A. This is a similar relationship but with the fecal coliform indicator bacteria and again showing a similar trend, again a highly significant correlation of .0001.
Q. And does it tell us anything with regard to the relationship between the fecal coliform and poultry waste?
A. So as fecal coliform numbers tend to be high, so does the concentration of the biomarker and vice versa, as they tend to be low, the concentration of the biomarker tends to be low. So they are correlated, they tend to co-vary.

Q. Does that mean the poultry waste biomarker co-varies with the indicator bacteria?
A. Correct.
Q. What is the chance of, let's say, a mistake in this analysis?
A. That would be, again, it's P less than .0001, so less than one in a thousand that this relationship occurred by chance.
Q. Now, Dr. Harwood, earlier I believe you stated an opinion concerning the importance of poultry waste as a contaminant, a bacterial contaminant in the IRW?
A. Correct.
Q. Would you please restate that opinion?
A. Yes, my opinion is that the poultry waste -- land application of poultry waste in the IRW is a major contributor to elevated indicator bacteria loads in the Illinois River Watershed in these waters.
Q. Now, what evidence did you use to reach this conclusion?
A. I used the weight of evidence approach which is what typically one does when investigating ecological questions. So rather than relying on one line of investigation, integrated numerous lines. So that would be starting out with -- and not in any particular order. But since we're talking about it, the widespread and quantifiable presence of the poultry litter biomarker and the evident pathway in terms of its concentration gradient from the litter to the fields to the edge of the field

improvement in water quality and a reduction in risk to human health.

MR. PAGE: Thank you. Your Honor, I pass the witness.

THE COURT: Cross-examination.

CROSS-EXAMINATION

BY MR. JORGENSEN:

Q. Dr. Harwood, how are you? Good to see you again.

A. Hello.

Q. We have met twice before; right?

A. Twice?

Q. Once at your deposition and once on the plane home.

A. Right.

Q. I think we were both very glad to be on the plane home.

A. Yes.

Q. So, Dr. Harwood, I believe you just stated your ultimate conclusion in this case and I want to say it again to make sure whether I get it right. Your conclusion is that the types of bacteria in the environment and the volume of bacteria in the environment in the Illinois River Watershed are likely from poultry litter?

A. A major source is from poultry litter.

Q. I'd like to show you a document that has previously been marked as Defendants' Exhibit 275 and ask you if you've seen it. Can you see that on your screen?

A. Yes.

MR. JORGENSEN: Let me give copies to the Court.

THE COURT: I have a copy of this.

Q. (By Mr. Jorgensen) Now, we're starting there on -- looks like there's some scribbles on the first few pages. So we're starting there on what is page 6. Have you seen that page before?

A. I don't recall.

Q. What does it look like to you?

A. It's a fax from Roger Olsen -- to Roger Olsen from David Page.

Q. To Roger Olsen from David Page. Who is Roger Olsen?

A. Roger Olsen is a -- I've been working with Roger Olsen on this poultry litter biomarker project and in general on the microbial sampling.

Q. And who is David Page?

A. David Page is the lawyer that was just asking me questions.

Q. Okay. And what's the date on this?

A. September 14th, 2005.

Q. Thank you so much. Let's turn to what in the exhibit is page 10 but -- and not 8, but 10. But on the numbers at the bottom of the page, it's 4 if you are following along on paper. I'll ask you to look at the paragraph labeled J there, source of bacteria. Let me read it and then ask you if that's right. Source of bacteria, Dr. Jodi --

THE COURT: Before we read it, once again in an abundance of caution here, this has already been referenced, but it is subject to the earlier stipulation between Mr. Bullock and Mr. George; correct?

MR. BULLOCK: Yes, it is, Your Honor.

MR. GEORGE: Yes, it is.

THE COURT: Very well, PI 275 is admitted.

MR. JORGENSEN: Thank you, Your Honor.

Q. (By Mr. Jorgensen) Let's look at this again. Do you see it there on your screen?

A. Yes.

Q. "Source of bacteria. Dr. Jodi Harwood will testify that the types and volume of bacteria in the environment is likely from land applied poultry waste and viruses associated with it." Let's scroll down just a little bit. PCR analysis may be used if we obtain poultry manure samples. Did I read that correctly?

A. Yes.

Q. When did you begin your work in this case?

A. April 2005.

Q. And when did you come to your conclusion?

A. Which part of my conclusion?

Q. The conclusion that --

A. The entire conclusion?

Q. Yes.

A. Really from the whole thing that I just described, it would have been late in 2007. Yes, late in 2007, because that's after we had analyzed the environmental samples with the biomarker.

Q. Did you know before today that Mr. Page had said this would be your conclusion before you ever even finished your work?

A. I don't know that he said that that's my conclusion since it's taken out of context.

Q. How is it taken out of context?

A. All I can see is that little box.

Q. Feel free to read the page.

MR. BULLOCK: Does the witness have a copy of it, Jay?

THE COURT: I don't know.

MR. JORGENSEN: May I approach, Your Honor?

THE COURT: Yes.

Q. (By Mr. Jorgensen) Did I read that correctly, Dr. Harwood?

A. That little segment.

Q. Okay. If your lawyer wants to ask you more questions about that, I'll let him do that, but the Judge limits us on time, so I'm going to move on. Your testimony is quite complex, so I'm going to try to simplify it and try to explain it. So let's start by talking about your role in the case, let's talk about what you did and what you didn't do. Is that

Q. And elsewhere?
A. Yes. And Salmonella was identified in edge of field samples and enumerated.
Q. Really?
A. Yes.
Q. You don't agree that the State took 68 samples for soil and found none with Salmonella in them?
A. No, I wasn't talking about soil. I was talking about edge of field. But soil, that could well be. I don't disagree.
Q. So what the State did find was fecal indicator bacteria, that's right?
A. The State did find fecal indicator bacteria, yes.
Q. Let's bring up Defendants' Demonstrative 33, if we can. I think this might help lay out what we've been talking about. I think it's 32. I'm sorry to have used the wrong number, it's 32. Okay. So you talked about fate and transport, you did not do a fate and transport analysis in this case?
A. Correct.
Q. Okay. So let's talk about what fate and transport is. What do you see on your screen there?
A. Well, can I restate that for a second or can I please restate my answer?
Q. Sure.
A. We didn't do a specific fate and transport analysis, but we did construct our sampling regime so as to be able to assess

Q. It's very prevalent.
A. It's -- it is common in many areas and -- but it's certainly more associated with fecally contaminated areas.
Q. Okay. And it comes from many sources?
A. That's right.
Q. As a matter of fact, almost every animal who sheds feces sheds fecal indicator bacteria?
A. Correct.
Q. So in the field I believe you testified that -- well, let me back up. So generally speaking, a fate and transport analysis, it refers to the elements and attributes that affect a bacterium's survival rate in the environment and the speed and manner with which it moves; is that right?
A. Those are some of the parameters that one investigates.
Q. Okay. So in a traditional fate and transport analysis, you're trying to see if something gets from point a to point B and how it might get there?
A. Yes, simplistically put.
Q. And it's much more important to do fate and transport or to understand that kind of a process where you have multiple sources of the item that you are looking for?
A. Can you ask me that question a different way? I'm not sure I follow.
Q. Sure. Isn't fate and transport that much more complex when the items that you're studying, the bacteria that you are

studying come from multiple sources?
A. Well, it really would depend on your study design. I can't say that. It depends on the question that you're asking.
Q. Is it easier for you to track one bacteria through the environment or multiple bacteria?
A. Multiple species, you mean?
Q. Yeah.
A. It would be easier to track one species than multiple species.
Q. And if the one type of bacteria comes from just one source, would it be easier to track it through the environment?
A. Compared to?
Q. Multiple sources.
A. Compared to a bacteria that comes from multiple sources?
Q. Exactly right.
A. Well, again, it would depend on the experiment design. It would depend on where you were starting and where you were ending up.
Q. All right. Well, let's move into those factors. Different bacteria move through the environment at different rates, don't they?
A. I'm not aware of any definitive research on that subject. It's pretty -- it's pretty well understood that many factors affect bacterial fate and transport, but it's not well understood how fast they move with respect to one another.

It's well understood, for example, that viruses move faster and farther than bacteria and that protozoa don't because viruses are small, bacteria are middle and protozoa are big.

Q. Different types of bacteria move through the environment at different rates; isn't that correct?

A. No, I don't -- I would not carte blanche agree with that statement.

Q. Do you remember giving a deposition in this case?

A. Yes.

Q. Do you remember that you were under oath when you gave that deposition?

A. Yes.

Q. Let's bring up, if we can, page 75, line 19 to page 76, line 2 in your deposition.

(An excerpt of the videotaped deposition of Valerie Harwood was played.)

Q. "Do you have an expert opinion on whether the types of bacteria in this case move at different rates?"

A. Did you ask me a question?

Q. (By Mr. Jorgensen) You're waiting to answer.

(An excerpt of the videotaped deposition of Valerie Harwood was played.)

A. "Bacteria move at different rates given the physical -- a lot of it has to do with the physical influences upon them and also has to do with their size. But so there are a lot of

factors that would influence whether they would -- at what rate they would move."

Q. (By Mr. Jorgensen) So to restate, bacteria move at different rates?

A. Depending on in part -- or in large part, I believe, on the physical and chemical factors that are influencing their movement.

Q. And those factors can include temperature?

A. For bacterial movement?

Q. Yes.

A. It could be a factor.

Q. Location within the water column?

A. Yeah.

Q. Presence of vegetation?

A. Yes.

Q. The media that they're moving through, whether it's grass or soil?

A. Yes.

Q. The size of the bacteria, some bacteria are big, some are small?

A. Again, the size differences don't make nearly as much of a difference as the physical and chemical factors.

Q. And the size of the spaces that they're moving through?

A. Correct.

Q. All of those are factors that affect how bacteria move?

A. Correct.
Q. So if you were to find a bacteria in the poultry house, you could not assume -- rather if you found two types of bacteria in the poultry house, you could not simply assume that they would move together?
A. If I found two types of bacteria in the poultry house and then what would happen to them?
Q. Could you assume that they would move through the environment together at the same rate?
A. Well, they're in the poultry house now, where are they going to go after that?
Q. If you found two different types, two different species of bacteria in a field, could you assume that they would move at the same rates?
A. I wouldn't want to assume it, I would want to test it.
Q. Okay. I think that's right. Bacteria also die at different rates; isn't that right?
A. Correct.
Q. A lot of factors affect how long they can survive out in the environment; right?
A. Correct.
Q. A bacterium's ability to survive depends on its own unique genetics?
A. Yes, and to the -- of course, the physical, chemical insults that it's subjected to.

Q. I think that's very important, so let's address those. So, for instance, in a field, a bacterium could be affected in its die-off rates by sunshine, oxygen, temperature changes, humidity changes, pH changes, salinity changes, predation changes and time?

A. Correct.

Q. All those things would kill bacteria at different rates?

A. Kill or inactivate or make non-viable.

Q. And a moment ago I believe you said that sunlight typically kills bacteria if it can reach the bacteria within two hours. Do you remember saying that?

A. Well, no, I didn't say if it would reach the bacteria within two hours. I said it would kill it within a couple of hours, that's a broad estimate, if the bacteria were directly exposed.

Q. Were directly exposed. So if I can use an example, in a cow pie -- this is kind of an embarrassing case and I'm just going to launch ahead.

A. Not to me.

Q. A cow pie is a little pie with a crust. Isn't it true that the bacteria inside that cow pie are protected from the sunlight or at least partially protected?

A. Yeah, yes.

Q. So they would die off at a much slower rate --

A. Than what?

Q. -- than if they were spread out on a field?
A. Correct.
Q. And if you were to spread out bacteria on the field in a thin, fine dust and thereby expose them to sunlight, those would die within a few hours?
A. Well, that depends on what you mean by a thin, fine dust.
Q. Thin enough that they could see the sunlight, they could be exposed to the sunlight?
A. If they are directly exposed, then they -- we're going to have a pretty high inactivation rate as long as they don't make it into the soil. If they do make it into the soil, then they'll be protected.
Q. And in talking about those same factors, dryness kills bacteria. I believe you used the word desiccation by that, but you mean dryness; right?
A. Correct.
Q. And that kills bacteria?
A. Correct.
Q. So the same thing, a cow pie shelters bacteria by keeping in the moisture; is that right?
A. Compared to?
Q. Compared to a thin dust?
A. Yeah, compared to a thin dust.
Q. Now, you're not offering an opinion in this case as to the relative rates of movement of bacteria that you've studied and

testified about; is that right?
A. Not to the relative rates of movement, no.
Q. In fact, as part of your work in this case, you did not study the movement characteristics of any type of bacteria in the watershed, did you?
A. No, I did not.
Q. Nor are you offering any opinion today about the different survival rates of the different bacteria in the Illinois River Watershed?
A. Can you rephrase that, sorry.
Q. Are you offering any opinion today as to the relative survival rates of the bacteria that you found in the watershed?
A. No.
Q. And you didn't study under what conditions and how long bacteria survived in this watershed, did you?
A. No, but we have done extensive studies of that in my lab.
Q. But you didn't study it here in the watershed?
A. Not in the watershed, no.
Q. Now, let's focus on the barn there on the screen. I've got that up as a representative of a poultry house. You don't know very much about the survivability of bacteria in poultry litter lying on a poultry house floor, do you?
A. I know that they're in a relatively stressful situation in that environment but I think you said relative survivability?
Q. Right.

A. Meaning with respect to one another?
Q. To each other, to one another.
A. We know that Enterococci tend to survive better than E. coli in poultry litter. That's one thing that's fairly well-established in the literature.
Q. And you know that poultry litter in houses is often layered, multiple layers go in?
A. Yes.
Q. And it sits there for a while?
A. Yes.
Q. Do you have an opinion whether the time that passes and the layering kills off the bacteria?
A. I would -- my opinion would be that -- which I haven't tested as we've established, but my opinion would be that the bacteria on the top layer of litter -- there are probably more viable and culturable bacteria on the top layer of the litter than there are at lower layers.
Q. And the ones at the lower layers would be dead or dying?
A. Well, they would be stressed at least.
Q. So you didn't study how long bacteria can survive laying out in a field after they were removed from a poultry house, did you?
A. Not specifically.
Q. You didn't study the specific fate and transport characteristics of bacteria moving between fields in the

watershed, did you?
A. No, I did not.
Q. And you didn't study the bacterial survival characteristics in the streams in the IRW?
A. Not specifically in the streams. Although again, we've done a lot of work in my labs, so I have a strong basis for opinions about that.
Q. You're not offering an opinion in this case as to the relative bacterial survival characteristics in the streams, are you?
A. You'd have to be a little bit more specific in your question.
Q. Did you study bacterial survival characteristics in the streams in the Illinois River Watershed?
A. Not in terms of an experimental study, no.
Q. All right. Let's walk through this demonstrative. So in a traditional fate and transport, you start in the poultry house, you move to the field where the litter is applied. And then you have to track how the litter moves, if at all, how bacteria in the litter move, if at all, as they encounter an edge of a field; is that right?
A. Well, there's all sorts of ways that you can design a study like that.
Q. Is that one way --
A. It depends on your questions.

Q. Is that one way to design it?
A. That is one way to design it.
Q. Then at the edge of a field you might encounter another field; is that right?
A. The edge of a field would be the edge, there would be something there to stop it.
Q. There would be something there to stop the bacteria from moving off the edge of the field?
A. No, there would be -- an edge of a field means an edge. There's something else there, a road, a ditch, something.
Q. Or another field?
A. I'd call that the same field.
Q. Okay. So it's your testimony that in the Illinois River Watershed all fields end in either a road or a ditch?
A. My concept of the term -- I'm sorry. Can I explain just briefly? My concept of what an edge of field is, is it's the end of a large, grassy expanse that would make up a field and then there would be something that would interrupt that grassy expanse, whether it be a ditch or a ditch and a road or a structure or something.
Q. And did you observe the sampling in this case?
A. No, I did not.
Q. So do you know if at the edge of the field, there was simply another field or always a ditch or a road?
A. In the edge of field samples that were collected in this

case, there was some sort of a ditch or a depression in which water could collect because those were water samples, the edge of field samples.

Q. So there were never -- if other witnesses have testified that there were puddles at the edge of a field, you contradict them?

A. No, I said a depression or a ditch or something where they could collect the water.

Q. In fact, you don't know what was at the edge of the field; isn't that right?

A. From what I've been informed, it's usually a ditch.

Q. In cases where it's a ditch or not a ditch, if there's another field beyond it, let's move through that, and then let's move through the demonstrative, and eventually then you reach the stream. If the question you are trying to address in a traditional fate and transport, and this is what I'm trying to bring out, that the bacteria in the stream came from the poultry house, don't you have to track it across the environment?

A. To demonstrate what?

Q. If you are trying to show --

MR. JORGENSEN: Your Honor, may I approach the demonstrative? It might help. We're having some trouble, maybe I can cut it short.

THE COURT: Yes.

Q. (By Mr. Jorgensen) Was the question that you were trying to address in this case, Dr. Harwood, whether bacteria that are found in the streams, whether those came from poultry litter? Is that the question you were trying to address?

A. Not directly whether bacteria that came from one particular field were in one particular stream, but whether there was a gradient of these signals from one compartment, in other words, from one type of sampling entity to another.

Q. So the bacteria that you find in a stream, E. coli, let's take that for example, they could come from cattle; right?

A. In certain streams there would be some possibility for contamination from cattle.

Q. They could come from birds?

A. There could be a bird component.

Q. If you found Salmonella, it could come from reptiles?

A. Salmonella has been isolated from reptiles.

Q. So if you found Salmonella in the streams of the Illinois River Watershed, it could come from reptiles? I'm not trying to trick you with these questions. I'm actually trying to clarify what you did.

A. So if I found Salmonella at an edge of the field sample I would --

Q. If you found Salmonella in the streams of the Illinois River Watershed, they could come from reptiles?

A. They could come from other sources other than -- than that

field, yes.
Q. And it was your job to help the plaintiffs understand whether the bacteria that you found in water, groundwater or streams, whether it came from poultry litter?
A. It was my job to determine whether or not there's a correlation between the practices of land applying this poultry litter and the contamination that's appearing in streams, that's how I would phrase it.
Q. And you did not do that through a traditional fate and transport analysis, you did it through the microbial source tracking we were just talking about?
A. We did the microbial source tracking, yes, as a way of determining whether or not we had a specific poultry litter signature in that water.
Q. All right. Now, let's talk for just a moment about the animals that live in the Illinois River Watershed. Pigs carry Campylobacter; is that true?
A. Pigs are not well-known to carry Campylobacter. I'm sure there's been a couple of studies that have found them.
Q. And Salmonella also, don't pigs also carry Salmonella?
A. Yes, pigs carry Salmonella.
Q. Most reptiles, I think we established, carry Salmonella?
A. I wouldn't say most reptiles, but I know they've been isolated from some.
Q. Humans contribute fecal matter to the Illinois River

Watershed directly?
A. Hopefully not.
Q. You don't know whether they contribute it directly?
A. No, I don't know.
Q. Let's look at page 186, line 14 of your deposition. Page 186, lines 14 to 21.
(An excerpt of the videotaped deposition of Valerie Harwood was played.)
Q. "So humans can contribute fecal bacteria to waterways directly?
A. "Directly, yeah, and also through their waste disposal systems.
Q. "Okay. And are septic systems a potential source of fecal pathogen contamination?
A. "Septic systems can be if they're not properly constructed to be separated from the water table."
Q. (By Mr. Jorgensen) Dr. Harwood, you haven't studied how many species of animals live in the watershed, have you?
A. No.
Q. You don't know how many types of birds live in the watershed?
A. No.
Q. You haven't studied the migration patterns of birds through the watershed?
A. Not directly, no. I've had some information on it, but I

have not myself studied that.
Q. You did not quantify the volume of manure deposited by each different type of animal in the watershed, did you?
A. Not myself, no. Although I have seen information on the subject again and I know that annually in the Illinois River Watershed there's about 350,000 tons of poultry litter land applied. I know that from Chris Teaf's work, that the volume of, for example, poultry litter is one of the dominant sources of fecal material contributed.
Q. Let's look at page 72, 19 of your deposition, 72, 19 to 21.
(An excerpt of the videotaped deposition of Valerie Harwood was played.)
Q. "Did you attempt to quantify the type of manure from each type of animal in the watershed?
A. No, I did not."
MR. JORGENSEN: And Then let's go to page 121, line 25 to 122, 2 of your deposition.
(An excerpt of the videotaped deposition of Valerie Harwood was played.)
Q. "Do you know the per capita fecal production of any living animal in the IRW?
A. "No."
MR. JORGENSEN: And then let's go to page 72, line 25 to page 73, 3.

(An excerpt of the videotaped deposition of Valerie Harwood was played.)

Q. "Did you attempt to quantify the volume of bacteria that come from each type of animal in the watershed?

A. "No, I did not."

MR. PAGE: Your Honor, I object to that use of the deposition. Her testimony was not that she tried to do it, but that she reviewed other people's materials, and that deposition statement there did not contradict her statements.

THE COURT: The question on the record that Mr. Jorgensen asked, I thought had to do with an attempt to quantify the type of manure. Just one second.

MR. PAGE: I believe the question, if I heard it correctly was, did she attempt to quantify it.

THE COURT: You have not determined the volume of manure deposited by each type -- I can't make it out -- of the watershed.

MR. JORGENSEN: I'm actually reading from a little script. So it's, "You did not attempt to quantify the volume of manure deposited by each type of animal in the watershed, did you?" And then the direct response is 72, Lines 19 to 21.

THE COURT: Overruled.

Q. (By Mr. Jorgensen) Dr. Harwood, did you attempt to quantify the volume of bacteria deposited by pets in the watershed?

A. No.
Q. Did you attempt to quantify the volume of bacteria, I'm not talking about the manure, but the bacteria in the manure deposited by humans in the watershed?
A. No.
Q. And you don't know whether anyone else on the State's team did any of these things, do you?
A. There was -- material was reviewed as to the relative or the amounts of animal feces that would be deposited in or that could contribute to impairments in the watershed, but that material -- that research was not done by me.
Q. And you're talking about the amounts of feces, not the volume of bacteria in the feces?
A. Correct.
Q. You didn't study the effects of urban runoff on bacterial loading in the watershed, did you?
A. No.
Q. All right. We've covered the things that you did and that you didn't do. Let's move to the science of microbial source tracking generally. Now, microbial source tracking, it's a young science; is that right?
A. I would say it started in 1996 or so, depending on where you start, so, yeah, it's 20 years old.
Q. Would you agree that it's still developing?
A. Yes, much as all of microbiology is developing.

important and where emerging methods are also important as long as they're based on reliable methods and good scientific validation.

Q. And in this case you've excluded work that was not based on a standard method?

A. Results, you mean, data?

Q. Uh-huh.

A. Yes.

Q. And in this case, the specific science that you are offering, the specific work that you did, it's novel, isn't it?

A. The work that I did is based on a technique that is validated reliable in many, many different fields. There are aspects of uniqueness to our approach, yes, but again, it's based on sound science and good validation.

Q. The question, Dr. Harwood, is the specific science that you are offering in this case, is it novel?

A. I don't know if I would use the term novel. It makes it sound kind of silly, but I would say it is a development of a new methodology. That's what I would say.

Q. It's untested, isn't it?

A. We tested it.

Q. It's not a standard analytical procedure?

A. It's not a standard analytical procedure.

Q. It's more appropriately considered developmental and cutting edge?

A. It is indeed, as I said, new. It is new method development.
Q. So no one else has done this before?
A. Other people have done very similar studies. Again, the EPA's own scientists are working on this methodology. They have peer reviewed publications out. It's not something that nobody has ever done before. It's not speculative. It's based on a reliable method and strong validation procedures.
Q. I believe you said a moment ago that it's not novel. Can we bring up Defendants' Exhibit 293? We start on page 2 of this at the very bottom. I think we need to give some context to this, otherwise it doesn't make sense and we want it to be fair. Does this begin with an e-mail from Roger Olsen to various people, including you?
A. Yes, it does.
Q. And does he say, "We are proposing to release all analytical data to the defendants. However, we don't want to release any of the PCR molecular tracking results at the time. Would the following statement preclude the PCR results?" And the statement is, "We will deliver to defendants copies of all chemical and bacteriological analytical results produced by standard analytical procedures and received from commercial labs, excluding any direct expert directed assessment manipulation, evaluation and our interpretation and opinions of the analytical results from all media, litter, soil

groundwater, surface water, lakes, rivers, streams, creeks and sediments."

All right. Let's go up to the next. That's a little bit of context. Let's go up to the next one, I think that might be on page 1. Is that an e-mail from Kent Sorenson to Roger Olsen?

A. Yes, it is.

Q. Let me read what Mr. Sorenson says. "Roger, to me it comes down to your definition of standard analytical procedures. While one could argue about whether the PCR or other techniques might be considered standard, I think we would be justified in saying this stuff is not standard, given that we're dealing with a potential biomarker that has not previously been demonstrated and for which we had to design new primers. In that sense, this is uncharted territory."

Did I read that right?

A. Yes.

Q. And then let's go to the e-mail above. Who is that from and to?

A. From Tanzem McBeth to Kent Sorenson, Roger Olsen and me.

Q. Does Tanzem say, "I agree with Kent, while the PCR itself may be standard, the process of developing the biomarker procedure is not standard. In fact, we haven't even finished developing and verifying the analysis and I think any disclosure of results at this point is premature"?

A. That was 2006.
Q. Let me go down to the last sentence. "The entire process is highly specialized and more appropriately considered developmental and cutting edge rather than standard."
Did I read that right?
A. Yes.
Q. And then the e-mail at the very top, who sent that?
A. That's from me to -- oh.
Q. Would you read what you said?
A. "I agree with Tanzem and Kent. This is method development in a relatively novel research area. Nothing is standard about it."
Q. Now, what you identified in this case is a bacteria, is that right? The biomarker that you refer to is a bacteria?
A. It's a gene from a bacterium.
Q. And it's not part of a chicken's DNA, I want to make that clear; is that right?
A. That's correct.
Q. It's not part of a turkey's DNA?
A. That's correct.
Q. It is a bacteria?
A. That's correct.
Q. And it's your theory that this bacteria lives in chickens and turkeys; is that right?
A. It's not a theory.

Q. Okay. So it can -- when it's found in the environment, it could be growing there on its own?
A. Now when it's found in the environment, that I don't know. But I know -- I strongly suspect that it could be cultured, so that would be growing outside of its host. But I don't know whether it could grow in the environment or not.
Q. Let's talk about whether this new bacterium is host specific. What does host specificity mean?
A. Host specificity is one of those funny words in microbiology. A lot of times I'd rather use the word host associated because almost any microorganism that you see can be found at a relatively low rate in some other organism. So host specificity would mean a strong -- in my mind host specificity means a strong association with a particular type of animal, animal species or a group of animals that one could define. So we'd find it much more frequently and at higher concentration in that organism than you would in other organisms, but I don't think of it as an absolute term.
Q. So host specific can mean -- or, well, let me say host specific does mean that it's specific to one type of animal?
A. So host specific, again the way that it's used in the literature means that it's predominantly found in one particular type of animal.
Q. You yourself have said that host specificity is the Holy Grail of microbial source tracking; is that right?

A. I wrote that, yeah.
Q. And host specificity is what a truly host -- host specific marker is what you're searching for in microbial source tracking; is that right?
A. Right.
Q. Because if it's not host specific when you find the bacterium, it could have come from multiple hosts; right?
A. If it's not host -- I assume you are using the term meaning absolutely host specific is how you --
Q. Right, if it's not absolutely host specific?
A. If it's not absolutely host specific, which most of the markers that we use in these studies are not, then you have to weigh the caveats of what other animals might be contributing and at what levels they might be contributing to the finding. And again, we're using the weight of evidence approach, so we're never relying solely on one angle, one line of evidence.
Q. So my question was if a bacterium is not host specific, then when you find it in the environment, it could have come from multiple hosts?
A. It depends on how many other hosts you might find it in, but it could have come from any sort of cross-reactive host that you find it in. Again, you have to weigh the lines of evidence.
Q. The marker, the biomarker in this case you've identified, it's not, in fact, unique to poultry, is it?

A. The biomarker that we identified is not unique to poultry. We found it in one duck sample out of the 10 that we analyzed and one goose sample out of the 10 that we analyzed. So it certainly meets the definition of strongly host associated but in terms of absolute host specificity, then it doesn't. So we have to be aware of that.

Q. So when you find this in the environment, it could have come from geese?

A. It -- if you find it in the environment in the absence of any other lines of evidence, then you wouldn't know whether it came from geese or not. Again, so you have to weigh everything in it.

Q. And the same for ducks?

A. Yes.

Q. And when you say you found it in one out of 10 samples, the one sample actually had the feces of 10 animals in it; right?

A. Right.

Q. So as far as you know, it could be in 10 ducks?

A. It was a very faint signal. And we actually used nested PCR to pick it up, rather than the qPCR, which is very, very, very sensitive and it was a very weak signal even then. And again, we tried to clone it and found it in very few of our clones. So we strongly suspect that it's at a very low level in these animals and probably in very few animals. But we

Q. You found it in ducks and geese?
A. One out of 10 samples, correct.
Q. Let's go to what is pages 8 and 9 of this exhibit. Did you test, Doctor, to know whether your bacterium is present in herons?
A. Herons?
Q. Uh-huh.
A. No.
Q. Coots?
A. No.
Q. Crows?
A. No.
Q. Hawks?
A. No.
Q. Owls?
A. No.
Q. Deer?
A. No.
Q. Any type of other bird?
A. No.
Q. Let's look down this list. Let's go to page 9. Do you see this long list of over -- I believe it's over a hundred different animals that live in the Illinois River Watershed, different types of animals that live in the Illinois River Watershed?

A. Yes.
Q. Did you test to see if your bacterium is present in any of those?
A. Nope, but can I explain something, Your Honor?
THE COURT: Yes.
THE WITNESS: When we determine which non-target samples or other animals to validate against, we target -- we choose the ones that are most likely to impact the watershed based on our knowledge of the watershed. Now, small birds, like many of these here, they have small masses of feces and their feces dry out quickly. Same with many -- most some animals. They simply aren't going to contribute a large microbial load to the water. So we -- it's impossible to go out and sample from all of these animals, so again we target the ones that, to the best of our knowledge, are going to be the major contributors to contamination throughout the watershed.
THE COURT: You've already made that point twice before; right?
THE WITNESS: Okay. Thank you.
Q. (By Mr. Jorgensen) I'll move on. Do you remember testifying that in this case you did not attempt to quantify the amount of feces or bacteria from any of these animals?
A. That's correct.
Q. Okay. Having identified this DNA sequence in an unknown

there been any foundation established that this witness has even seen this document before or is part of a correspondence chain or anything?

THE COURT: Sustained.

MR. JORGENSEN: I'm sorry, what's that?

THE COURT: Sustained.

Q. (By Mr. Jorgensen) Have you seen this before?

A. No.

Q. Do you agree with the assertion that your method is so new as to be proprietary?

A. I don't know.

Q. It is new, isn't it, and unlike what has been done before?

THE COURT: I think we've plowed this ground before. Let's take a break. We'll take a five to ten minute recess.

(Recess.)

Q. (By Mr. Jorgensen) Dr. Harwood, in this case you did not personally gather any of the samples that you analyzed, did you?

A. That's correct.

Q. But the samples that were provided to you, there were samples from ten cattle fields; is that right?

A. Yes, that's right.

Q. If I left this building and went and found ten cattle fields in the neighborhood and none of these cattle in those fields had trichinosis, does that mean that none of the cattle

in Oklahoma have trichinosis?
A. No.
Q. Can we bring up what we previously showed, as I believe you called it a cartoon, Defendants' Demonstrative Exhibit 32. Now, Dr. Harwood, because you did not study the fate and transport of the new bacterium, you do not know whether, if it were in a poultry litter house or on a poultry litter field, whether it would move in the same manner and at the same rate as other bacteria?
A. I have no reason to believe that it wouldn't.
Q. Aren't bacteria of -- I think we've established this. Aren't bacteria of different types -- don't they move differently?
A. I didn't agree with that. I said that the physical and chemical factors that influence them are going to be more important than their type.
Q. So you do not agree that some bacteria are large and some are small?
A. Some are large and some are small, but within a very -- I mean, over an order of magnitude.
Q. Some move quickly and some don't, you don't agree with that?
A. Their actual movement, their motility is not going to be nearly as important as the physical forces that are moving them.

Q. Do you recall that you warned the Court in that case about enteropathogenic E. coli and the symptoms of it?
A. Yes.
Q. Did you not say that those symptoms could result in kidney damage or death?
A. Yes.
Q. Then you specifically reference the O157:H7 strain?
A. Yes.
Q. You did not test -- let me restate that. The plaintiffs did not test anywhere in the watershed for 0157, did they?
A. That's correct.
Q. You have no evidence that 0157 exists in the watershed?
A. Not from our study.
Q. And the 0157 Enterococcus is associated with healthy cattle; is that right?
A. You mean 0157 E. coli?
Q. Yes, 0157:H7 E. Coli associated with healthy cattle?
A. It can definitely be isolated from healthy cattle, yes, and from cows and other animals.
Q. When you warned the Court about kidney damage or death for a bacteria for which you did not test, were you trying to scare the Court?
A. No, I'm sure the Judge -- Your Honor, you've seen my affidavit and it simply is a list basically of some pathogens that one might find associated with poultry feces. Nowhere in